

## Insects as an Alternative Source for the Production of Fats for Cosmetics

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### Synopsis

Insects may provide an environmentally friendly way of producing high-quality bio-based materials that can be implemented for cosmetic applications. Insects can be bred on organic waste, in high numbers, and on small surfaces, therefore, making large scale industrial breeding possible. Fats from three insect species: the black soldier fly (BSF) (*Hermetia illucens*), the locust (*Locusta migratoria*), and the house cricket (*Acheta domesticus*) were evaluated for potential use in skin care. Insects were dried and fats were extracted using petroleum ether. The fats were further refined, and the fatty acid composition and the acid value were determined. The fats were used in a hand cream formulation and compared with the currently used mink- and plant-derived oils. Fatty acid analysis indicates that BSF contains >60% of lauric acid, which makes it less suitable for application in a skin-care product, whereas locust and cricket fats are rich in C16 and C18 fatty acids which makes them more suitable. Phospholipids and free fatty acid levels in the three insect species are relatively high compared with commercial, refined oils, and need to be removed by appropriate refining protocols. Odor and color also need to be removed by physical refinement to improve the applicability.

### INTRODUCTION

In the modern industry, the development of novel materials and products with little impact on the environment and of no concern for human health is of crucial importance. The search for novel materials is not only driven by the decline in fossil resources that will eventually become limited but also by the impact that human activities have on the planet, e.g., in waste production and overexploitation of natural sources. As an alternative to fossil oil, the industry is increasingly using plant material as a source for oleochemical applications (1). Although at first sight this may seem an environmentally friendly source of oil production, its impact on the environment is considerable (2) as the increase in demand for vegetable oils and biofuels contributes to tropical deforestation, habitat

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fragmentation, and loss in biodiversity (3,4). Therefore, research into alternative sources of bio-based chemicals is needed.

Another large impact of human activities on our planet is the production of food and resulting associated waste, such as organic food-waste streams. Studies suggest that food production needs to increase dramatically to keep pace with projected demands from population growth, dietary change (meat use), and increasing bioenergy use (5,6). Although several potential solutions to these problems are being investigated, food production also generates large food waste streams that should be better exploited. If these wastes can be converted into profitable biomass, then the ecological burden that waste poses may decline.

Insects may contribute to the development of this circular economy, and they already find implementation in the food and feed sectors. Although insect consumption is common in large parts of the world, this is not an established practice in the western world. However, insect consumption may have a large impact on food/feed-associated problems, and therefore, work is done to demonstrate the potential of edible insects to the regulatory agencies (7).

Another application of insects can be as a source of biomaterials, such as fats, proteins, and chitin. These broad fractions of biomaterials may find diverse use in the industry (chemical). Coupling the breeding of the insects to waste stream reduction (8), insects may become a sustainable alternative in the production of biomaterials, such as fats and oils.

An example is the work being done on the black soldier fly (BSF, *Hermetia illucens*). BSF are relatively easy to breed in large quantities on small surfaces. The larvae can be easily bred on organic waste streams (9) and the resulting fats are being explored for their potential use in biofuels (10,11). Also chitin, extracted from BSF, can be modified into chitosan. This molecule has antimicrobial activity that may find application in several areas, including food and nutrition; biotechnology; material science; and pharmaceuticals (12).

Other insect species that are commonly investigated are the house cricket (*Acheta domestica*) and the locust (*Locusta migratoria*) (13). The house cricket is usually grown on pet food (fish food/cat food) under lab conditions, but they can also be bred on organic vegetable wastes. Locusts are bred on grass, which may pose an issue for breeding during winter in northern countries, but still, a case can be made to use locusts in the reduction of the large amounts of grass waste that are produced yearly with no relevant application.

Fats and oils are commonly used in cosmetics, where they are a major component of creams for skin care. Triglycerides typically act as emollients that soften the skin (14). Indirectly, they will also moisturize the skin by reducing the transepidermal water loss (TEWL) (15). Depending on the fatty acid profile, the properties of the fats can vary and the healing (e.g., using linoleic acid for dry skin) or skin-protective functions of the creams can be enhanced. The fats are also used to increase the viscosity of the formulation or for their emulsifying properties (16). From a skin-care point of view, mink-fats have a favorable fatty acid profile and have therefore been traditionally used in many skin-care formulations. Although the mink oil can be seen as a side stream derived from mink fur industry, ethical objections arise toward the use of minks for their fur. Other sources of useful fats need to be explored. Macadamia nut oil, which has a similar fatty acid profile as mink oil, has been shown to present a suitable source of oils for cosmetics. However, issues may arise with the use of edible oils cultivated on valuable land for nonfood/feed

applications and transport and associated costs. Therefore, identifying other suitable sources such as insect fats may prove useful.

In this paper, three insect species, the BSF, the house cricket and the locust, were evaluated for their potential use in cosmetic applications. Fats extracted from these insects are evaluated in a hand cream formulation as a proof of principle to demonstrate the potential of using insects, cultivated on organic waste streams, in personal health-care products.

## MATERIALS AND METHODS

### INSECTS

In general, insects are considered as farm animals and are to be treated as such according to the Belgian legislation (17). However, detailed guidance regarding insect welfare and euthanasia are lacking and little is known about the humane treatment and other ethical aspects in insects. Black soldier flies were bred at the Thomas More campus Geel in a greenhouse at an average day temperature of 30°C and relative humidity of 50–90%. The breeding process is similar to the process described in Sheppard et al.(18). Female flies deposit their eggs in cardboard structures. Eggs were harvested and placed shielded from light until they hatched. For this study, the larvae were cultivated on chicken feed. When they reached the pre-pupae stage, the pre-pupae migrated out of the chicken feed and were harvested. The pre-pupae were stored at –20°C until needed for fat extraction.

Locusts and crickets were bought frozen from a local insect breeder (Desmedt Insects, Tessenderlo, Belgium).

### CHEMICALS AND FATS

All chemicals, citric acid, sodium hydroxide, hexane, chloroform, propanol, diethyl ether, methanol acetic acid, phenolphthalein, Fuller's earth, ethyl acetate, petroleum ether (40–65°C), NaCl, BF<sub>3</sub>, 2,2,4-trimethyl heptane, Na<sub>2</sub>SO<sub>4</sub> used for the extraction, degumming, thin-layer chromatography (TLC), and gas chromatography–mass spectrometry (GC–MS) procedures were bought from Sigma-Aldrich (St. Louis, MO) and VWR Chemicals (Radnor, PA). Macadamia nut oil and a commercial vegetal oil mixture (a mixture of macadamia nut oil, cotton seed oil, and olive derived squalene) were purchased from IMCD Benelux BV (West-Knollendam, The Netherlands) and Clariant (Puget-sur-Argens CEDEX, France), respectively.

### EXTRACTION OF INSECT FATS

Fats were extracted from all three insect species. The insects were dried, typically overnight at 65°C, and subsequently ground and sieved (maze 2.36 mm).

A Soxhlet procedure using petroleum ether was applied to extract fats from small (15 g) insect samples. For preparation of larger volumes, the fats were extracted at room temperature in hexane. About 3 l of hexane was added to 1.5 kg of grinded insect material.

The solution was incubated while stirring for 1 h and was then filtered using an air press filter, separating the hexane/fat mixture from the remaining solid fraction (consisting mainly of chitin and proteins). The petroleum ether (Soxhlet extraction) or hexane (large scale extraction) was subsequently removed (and recycled) by evaporation in a rotavapor device and the yield of raw fat was determined.

#### FRACTIONATION OF BSF FATS

BSF fat was fractionated according to a modification of the method published by Kaluzny et al. (19). A 500-mg aminopropyl solid-phase extraction column (Chromabond NH<sub>2</sub>, Macherey-Nagel; column 1) was washed twice with 2 ml hexane. A 10 mg sample of raw BSF fat was dissolved in 0.5 ml CHCl<sub>3</sub> and applied to the solid-phase extraction column (SPE) column, and the CHCl<sub>3</sub> was discarded. Four milliliters of CHCl<sub>3</sub>:2-propanol (2:1) was applied on the column and the elute was collected in Tube 1. Subsequently, 4 ml of 2% acetic acid in diethyl ether was applied on column 1 and the elute, which contains the free fatty acid (FFA) fraction, was captured in a separate tube. Column 1 was then eluted with 4 ml methanol and the elute, which contains the phospholipids, was collected in a separate tube. SPE column 1 was then discarded. The eluent in Tube 1 was then blow-dried with N<sub>2</sub> gas and redissolved in 0.4 ml hexane. A second aminopropyl SPE column was prepared by washing it twice with 2 ml hexane and the redissolved eluent (Tube 1) was applied on the column. Four milliliters of hexane was applied. The eluent, which contains cholesteryl esters, was captured in a collection tube. Six milliliters of a 1% diethylether, 10% dichloromethane in hexane solution was applied and the eluent, which contains triglycerides and cholesterol, was collected. The SPE column was then treated with 6 ml 5% ethyl acetate in hexane and the eluent (containing cholesterol) was collected. Diglycerides were collected by eluting the column with 4 ml 15% in hexane. Monoglycerides were eluted by applying 4 ml CHCl<sub>3</sub>:methanol (2:1). All collected fractions were dried under N<sub>2</sub> gas and subsequently dissolved in 200 µl CHCl<sub>3</sub> for visualization using TLC.

#### TLC

TLC was run on a 20 × 20 cm silica gel plate (ALUGRAM SIL G/UV<sub>254</sub>; Macherey-Nagel, Duren, Germany). Samples were loaded as individual spots separated by 1 and 1.5 cm from the bottom of the plate. Samples were loaded using an individual spot for the fat samples (obtained by dissolving 50 µl fat in 0.5 ml CHCl<sub>3</sub>) and applying three spots for the samples that were obtained by fractionation of the BSF fats. One hundred and two milliliters of running buffer (80 ml hexane:20 ml diethylether:2 ml acetic acid) was poured in a glass container and the loaded TLC plate was put in the tank and run until the front reached the top of the plate. The TLC plate was then put in a glass tank for I<sub>2</sub>-vapor visualization.

#### REFINING OF INSECT FATS

Raw fats still contain contaminants, such as phospholipids and FFAs, which limit in the shelf life. These contaminants should be removed by a degumming process. The procedure described here starts from 50 g of fats, but adapted volumes can be taken for other

starting volumes of fat. Fifty g of fat was heated to 90°C and 15 ml of a 50 mM citric acid solution was added and incubated for 30 min (90°C) while stirring. The solution was centrifuged (5 min, 2,000 × g) and the oil phase was washed with 10 ml warm water followed by centrifugation (5 min, 2,000 × g) and retaining of the oil phase. The wash step was repeated once.

The degummed fat was heated to 95°C while stirring and 10% NaOH was added to neutralize the FFAs for 45 min. The mixture was centrifuged for 10 min (2,000 × g) and the solution was washed twice with 6 ml demi water.

Bleaching of the fats was done by adding 1% of Fuller's earth and incubating at 95°C for 30 min. The fat fraction was removed by centrifugation (5 min, 2,000 × g).

Finally the fats were deodorized by removal of the remaining solvents, aldehydes, ketones, and alcohols using a vacuum steam distillation, run for 3 h.

#### ACID VALUE

The acid value was determined by titration with phenolphthalein as an indicator. 0.25 g fat was dissolved in 50 ml ethanol and was titrated with a standardized NaOH solution. The acid value was subsequently calculated according to the formula [based on (20)]:

$$\frac{V \times C \times FW}{m \times 1,000} \times 100 \times 1.99 = \text{acid value}$$

Here  $V$  is the volume of NaOH added to reach the inflection point,  $C$  is the effective concentration of NaOH, and  $m$  is the mass of fat. FW is the approximate average molecular weight of the fatty acid profile of the insect (e.g., ±235 for BSF and ±275 for the cricket and the locust). 1.99 is a conversion factor.

#### FATTY ACID PROFILE DETERMINATION WITH GC-MS

The fatty acid composition was determined by GC coupled with MS using an Agilent 78020A.

The fatty acids were first converted to fatty acid methyl esters to make them volatile. 1.5 ml 0.5 M methanolic NaOH was added to 25 mg fat and the mixture was heated to 100°C for 5 min. The mixture was then cooled to room temperature, 2.4 ml of the catalyst BF<sub>3</sub> was added, and the mixture was incubated at 100°C for 30 min. The mixture was cooled to room temperature, 1 ml isoctane was added, and the mixture was vortexed. Five milliliter of a sodium chloride solution was added, the mixture was vortexed and subsequently centrifuged at 2,700 × g. The supernatant was collected and the pellet was washed twice with isoctane, followed by centrifugation and collection of the supernatant. The supernatant was dehydrated using Na<sub>2</sub>SO<sub>4</sub>, and after filtering, the solution was brought to 5 ml with isoctane. From this mixture, three dilutions (each containing an internal standard methyl heptadecanoate) in hexane were made for analysis by GC-MS. Quantification was performed by comparing the chromatogram to standard curves derived for each fatty acid that was to be detected.

## FOURIER TRANSFORM INFRARED SPECTROSCOPY

Infrared spectra of the fats were acquired on a Brüker ALPHA spectrometer (Brussels, Belgium) with a platinum ATR single reflection diamond module. Each spectrum was the result of averaging 24 scans taken at a resolution of  $4\text{ cm}^{-1}$ . All spectra were measured at  $21^\circ\text{C}$ .

## CHALLENGE TEST

The efficacy of microbial preservation was tested according to the method described in the European Pharmacopoeia version 7.0 (21). The inoculum consisted of *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Escherichia coli*, *Candida albicans*, and *Aspergillus brasiliensis*.

## VISCOSITY

The viscosities of the oils were determined using 125 ml of the oils in 250 ml beakers at  $25^\circ\text{C}$  using a Brookfield (Elscolab, Kruibeke, Belgium) type RVDV-II + programmable viscometer using spindle 3 at 60 rpm. Several other oils were measured as a reference. These were isopropyl myristate (BASF, Ludwigshafen, Germany), cyclopentasiloxane (Dow Corning Europe, Barry, UK), octyl stearate (BASF), C12–C15 alkyl benzoate (Innospec, Littleton, CO), decyl oleate (BASF), C8/C10 triglyceride (BASF), octyldodecanol (BASF), and dimethicone (BRB International, Ittervoort, The Netherlands).

## SPREADABILITY

The spreadability of the oils was measured following the protocol by Dietz (22). Gelatin films made from lime bone were kindly donated by Rousselot (Gent, Belgium). Five milliliter of oil was pipetted onto the films and after 5 min the drop diameter was measured using a caliper. The reported values are an average of 4 or 8 measurements.

## FORMULATING OF HAND CREAMS

The insect fats were formulated as 1%, 2%, 4%, 5%, and 10% fractions in a hand cream. The composition of the formulation and the origin of the used materials are given in Table I. The typical formulation contains 5% fat, and the variation in fat content was compensated by adding more or less water in phase 2. The formulations with insect fats were compared with formulations containing 5% mink or macadamia nut oil. Macadamia nut oil was purchased from IMCD Benelux BV. The origin of the mink oil is proprietary information. The natural antioxidant sunflower seed oil extract of rosemary leaf (INCI Helianthus Annuus Seed Oil Rosmarinus Officinalis Leaf Extract) was obtained from Gattefossé.

Table I  
Hand Cream Composition (The Fat % x Can Be 1, 2, 5 or 10 %)

	INCI	Function	%	Supplier
Phase 1	Glyceryl stearate	Emulsifier	2–3	Gattefossé
	Behenyl alcohol	Viscosity enhancer/stabilizer	2–3	BTC Europe
	Cetyl Phosphate	Emulsifier	1–2	DSM
	Cetearyl Ethylhexanoate	Emollient	2–3	Mosselman
	<i>Fat/oil</i>		x	—
Phase 2	Dimethicone	Emollient	1–2	BRB International
	Aqua		q.s.	—
	Acrylates/C10-30 Alkyl Acrylate Crosspolymer	Viscosity enhancer/stabilizer	0.3–0.5	Lubrizol
	Glycerin	Emollient/moisturizer	3–4	Vivochem
	Methylpropanediol	Emollient	3–4	Stearinerie Dubois
	Caprylyl Glycol	Wetting agent with strong antimicrobial properties	0.3–0.6	Dr Straetmans
	Disodium EDTA	Chelator	0.02	BTC Europe
	Aqua		5	—
Phase 3	Sodium Hydroxide	pH adjuster	0.6	Brenntag
Phase 4	Phenylpropanol	Fragrance ingredient with antimicrobial activity	0.1–0.2	Dr Straetmans

The hand cream was prepared according to the following protocol. All components of the various phases were weighed separately. Next, the oil and aqueous phases, phases 1 and 2 respectively, were heated to 75°C. Once this temperature was reached, phase 1 was gradually added to phase 2 while mixing with a Silverson mixer until a homogeneous cream was obtained. This cream was left to cool down to 30°C at which point phase 3 was added. The rise in pH due to the addition of 30% NaOH will cause the acrylates/C10-30 alkyl acrylate crosspolymer to thicken the cream. Finally, once the hand cream was at room temperature the fourth phase component, phenylpropanol, which serves together with methylpropanediol and caprylyl glycol as preservative, was added. The pH of the cream should be between pH 5 and 6. If not, the pH is adjusted using 30% NaOH solution or 50% citric acid solution.

#### STABILITY TESTING OF HAND CREAMS

Hand creams containing 1%, 2%, 4%, 5%, and 10% of insect fat were formulated and placed in incubators at 5°C and 45°C, and also kept at room temperature (21°C). At regular time intervals, up to 8 weeks, color, odor, and aspect were evaluated.

## RESULTS AND DISCUSSION

#### EXTRACTION AND REFINING OF INSECT FATS

After harvesting, the insects were dried at 65°C until constant weight before extraction. Starting from whole insects, the average percentage of fats to dry weight (regardless of

**Table II**  
Yield (Average Percentage) of Raw Fats Extracted from BSF, House Cricket and Locust

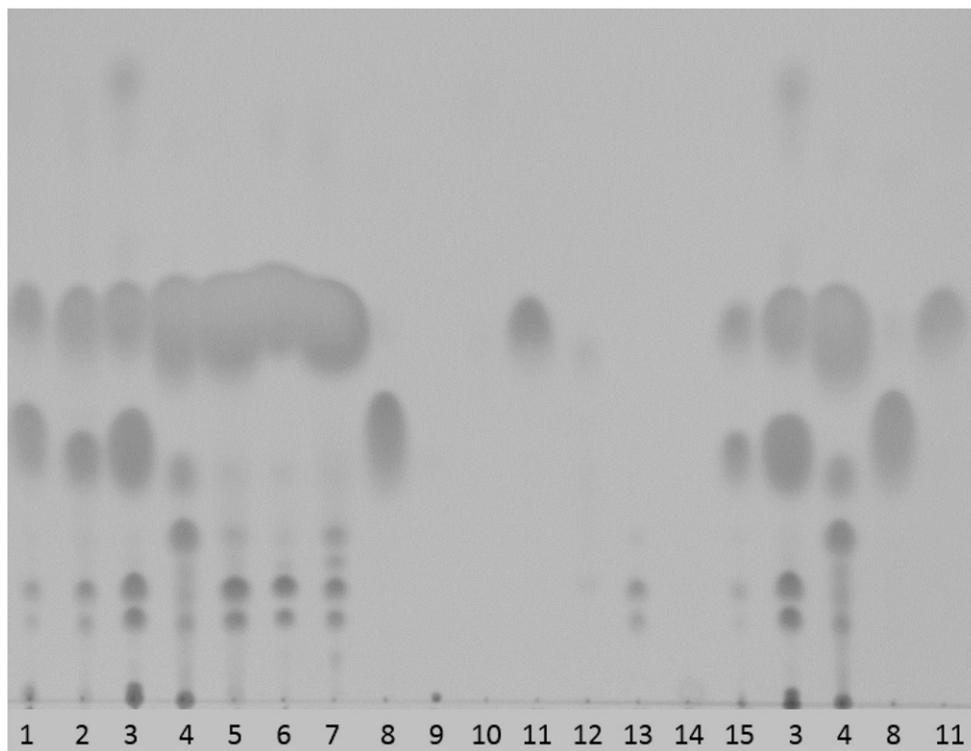
Species	Average % yield (Soxhlet)	Average % yield (large extraction)
BSF	33.9 (N = 9)	27.2 (N = 3)
House cricket	20 (N = 3)	15 (N = 2)
Locust	22.5 (N = 6)	17.3 (N = 1)

Soxhlet method versus large batch extraction.

the starting amount) is shown in Table II. The number of extractions that were performed is given within brackets. For locusts only, one large batch extraction on 1.787 kg dry material was performed.

#### FRACTIONATION OF FAT AND TLC ANALYSIS

A BSF sample was fractionated according to a slight modification of the protocol published by Kaluzny et al. (19). In Figure 1, a TLC image is shown in which fats of BSF, cricket, locust as well as other oils that are applied in cosmetics (mink oil; vegetal oil;



**Figure 1.** TLC of insect fats and fractions of BSF fat. Lanes: BSF (samples 1, 2, 15); cricket (3); locust (4); mink oil (5); macadamia nut oil (6); vegetal oil (7); fractionated BSF fractions: FFAs (8); phospholipids (9); cholesteryl esters (10); triglycerides (11); cholesterol (12); diglycerides (13); monoglycerides (14).

macadamia nut oil) and the fractionated BSF fractions (FFAs; phospholipids; cholesteryl esters; triglycerides; cholesterol; diglycerides; monoglycerides) are presented.

Comparison of the insect fats, mink oil, vegetal oil, and macadamia nut oil with the fat fractions indicates that BSF and cricket fats contain a rather large proportion of FFAs. In locust fat, the number of FFAs is lower. In the oils that are typically used in cosmetics (Figure 1, Lanes 5 and 6), the fraction of FFAs is limited but these oils have been thoroughly refined. Phospholipids (a spot close to the bottom of the TLC plate) can also be seen in the BSF, cricket, and locust fats and is almost absent in the refined fats. Fractions that are abundantly present in all oils are the triglycerides and the diglycerides. Cholesteryl esters and monoglycerides are hard to observe in the samples.

Refining of the raw fats (starting with 50–150 g of fat) encompassed a total degumming, neutralization, bleaching, and deodorization procedure. Bleaching and deodorization had only slight effects, but the fats became less dark and the odor less pronounced (data not shown). The locust fat had a greenish color that was hypothesized to be due to the presence of chlorophyll as the locusts were fed grass. This was verified spectrophotometrically as the UV–vis spectrum showed the typical absorption bands of chlorophyll (data not shown).

To evaluate the refining process and to characterize the fats, the acid values, which are a measure of the FFA content, were determined. The acid values of the fats are shown in Table III. These values are high compared with the acid values of the thoroughly refined mink oil (0.48 mg/g fat according to the manufacturer's specifications) and macadamia nut oil (0.13 mg/g fat according to the manufacturer's specifications). This indicates a significant presence of FFAs in the raw insect fat fraction. Although locusts still contain a considerable amount of about 20 mg of FFAs/g fat, this is far less than what is found in BSF and cricket fats. These values are corroborated by the Fourier-Transform Infrared Spectroscopy (FTIR) spectra (Figure 2), which show the presence of a significant peak around  $1,710\text{ cm}^{-1}$  in the case of the cricket fat and a far less intense peak in the case of locust fat. The peak around  $1,710\text{ cm}^{-1}$  can be attributed to the presence of the C=O stretching of the carboxyl group of FFAs, whereas the absorption band at  $1,743\text{ cm}^{-1}$  arises from the C=O stretching of the ester carbonyl group of the triglycerides (23). The FTIR data also illustrate the effect of refinement on decreasing the amount of FFAs within the three insect fats.

#### FATTY ACID PROFILE

The fatty acid profiles of the three insect species were determined by GC–MS and are presented in Table IV. The table also includes typical average values for mink oil and macadamia nut oil (information provided by suppliers). There is a large difference in fatty acid composition between BSF and the two other species. BSF fat stands out with its high

Table III  
Average Acid Value (mg FFA/g Fat) of Fats of three Insect Species

Species	Average acid value (Soxhlet)	Average acid value (large extraction)
BSF	91.6 (N = 2)	86.2 (N = 7)
House cricket	74.4 (N = 2)	68.1 (N = 5)
Locust	22.2 (N = 2)	20.7 (N = 3)

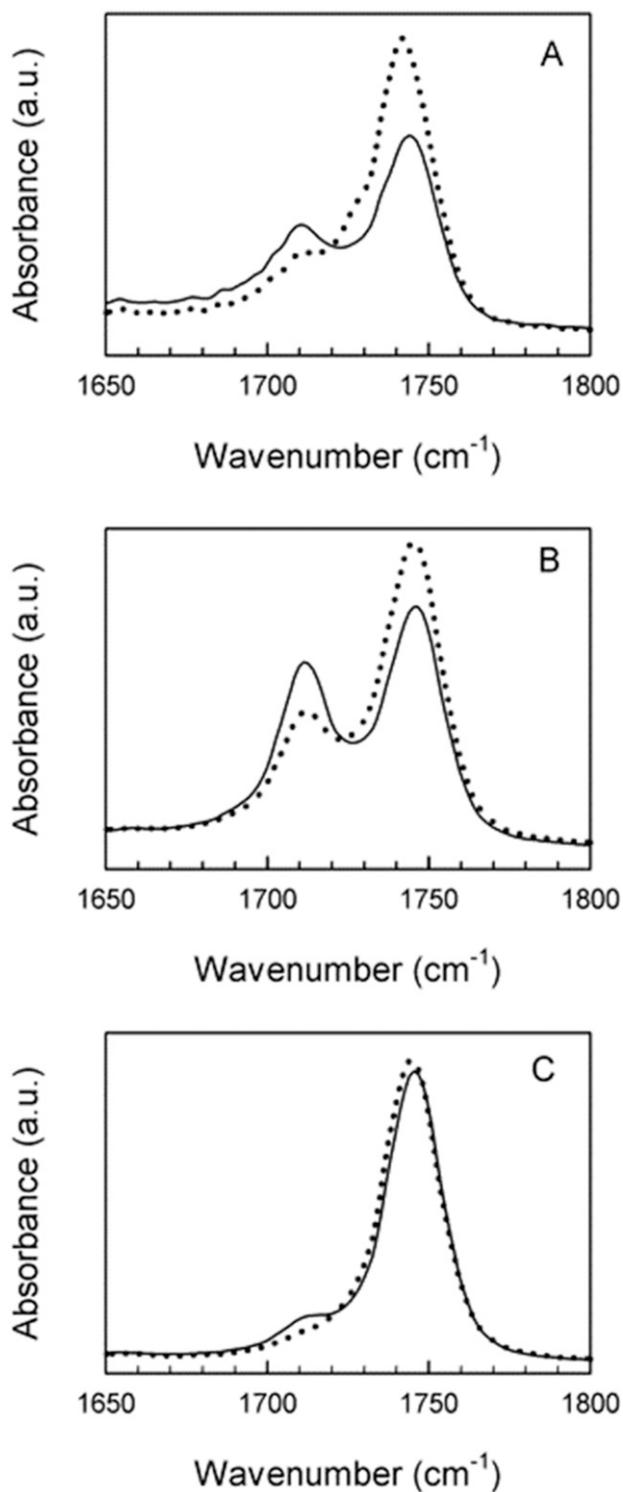


Figure 2. FTIR spectra (1,600–1,800  $\text{cm}^{-1}$ ) of BSF (A), cricket (B) and locust (C) fats raw fats solid line, refined fats dotted line).

Table IV  
Fatty Acid Profile in three Insect Species as Determined by GC-MS

Fatty acid methyl ester	Fatty acid-chain	BSF %	Cricket %	Locust %	Mink %	Macadamia nut %
Methyl decanoate	C10:0	<1	/	/	/	/
Methyl laurate	C12:0	58	<1	<1	<1	<1
Methyl myristate	C14:0	8	<1	2	4	<1
Methyl myristoleate	C14:1	/	<1	<1	<1	/
Methyl palmitate	C16:0	10	26	24	16	8
Methyl palmitoleate	C16:1	2	<1	<1	15	22
Methyl heptadecanoate	C17:0	<1	/	<1	<1	/
Methyl stearate	C18:0	1	11	12	3	3
Methyl oleate	C18:1	10	24	30	41	59
Methyl linoleate	C18:2	9	35	17	17	3
Methyl linolenate	C18:3	<1	2	14	1	/
Methyl arachidate	C20:0	<1	<1	<1	<1	2
Methyl eicosanoate	C20:1	/	/	/	/	2
Methyl 5,8,11,14,17 eicosapentanoate	C20:5	/	<1	<1	/	/
Methyl behenate	C22:0	/	/	/	<1	/
Methyl lignocerate	C24:0	/	/	/	<1	/

percentage of relatively short chain FA's (C12 and C14). It is also characterized by an extremely low degree of unsaturation (~21%) compared with ~61% for cricket and locust fats. By contrast, mink and macadamia nut oils contain 75.2% and 83% unsaturated fatty acids, respectively. All insect fats have a low percentage of palmitoleic acid (C16:1) compared with mink and macadamia nut oil. The palmitoleic acid is generally considered to be responsible for the good skin penetration properties of these oils (14). Both locust and cricket fats contain significant amounts of saturated and unsaturated C18 chains, comparable with the mink and macadamia nut oils. The relative abundance of stearic acid (C18:0), oleic acid (C18:1), linoleic acid (C18:2), and linolenic acid (C18:3) is consistent with previous findings on insect fats (14,24).

Based on the fatty acid composition, it is clear that cricket and locust fats are more suitable for use in cosmetics. Oils or fats rich in linoleic acid and linolenic acid are known to reduce the TEWL and regenerate the lipid barrier of the epidermis (24,25). The skin feel may not be ideal based on the low amount of palmitoleic acid present, but this is not necessarily an issue, depending on the type of application. The use of BSF fat should be avoided in leave-on products as its high lauric acid (C12:0) content is likely to cause adverse effects on the skin's lipid structure. It is likely that a large fraction of the observed FFAs is lauric acid, which—as a FFA—is known to disrupt the skin barrier and increase the TEWL (26). However, the fatty acid profile of the BSF fat is very similar to that of palm kernel oil and coconut oil. As such, BSF fat derivatives such as surfactants could find similar uses as palm kernel oil and coconut oil derivatives in rinse-off products, such as soaps and shower gels.

Note that the fatty acid composition of an organism depends on various factors, such as feed and life stage (24,27). The BSF fat is obtained from pre-pupae whereas the locust and cricket fats are extracted from adult animals. This may provide an explanation for the observed discrepancy in fatty acid composition. Liu et al. (27) have shown that large differences in fatty acid profile occur among the different life stages of the BSF. However, the

amount of lauric acid is high from the larvae stadium up to the adult fly, suggesting that the high C12-content is probably species specific.

#### SPREADABILITY AND VISCOSITY OF THE INSECT FATS

The appreciation of a cosmetic product not only depends on its functionality, for instance hydrating or anti-wrinkle properties, but also on the skin feel. Major factors in skin feel are the lubricity, spreadability, and perceived greasiness (14). The latter is related to the rate at which an oil or fat penetrates the skin. The fatty acid composition plays an important role in skin feel. As a general trend it is expected, for natural plant and animal derived oils, that the higher the degree of unsaturation, the lower the viscosity, and the higher the penetration rate will be. This will result in a less greasy skin feel. This is, however, just a trend line, as notable exceptions exist. Often these deviations from the trend line are related to the presence of an unsaponifiable fraction. Mink oil, for example, is such a deviant oil with a good skin penetration despite its relatively high viscosity.

The viscosity and spreadability of the insect fats were determined and compared with those of mink and macadamia nut oil (Tables V and VI). The viscosity of several other oils were measured as a control. A comparison of the values obtained in this work with those reported by Dietz (22) using a falling ball viscometer results in a correlation plot with  $r^2$ —value of 0.889. This suggests that the relative order of the viscosities of the oils is correct.

Both mink and macadamia nut oil, which have comparable fatty acid profiles, are characterized by a high viscosity and a low spreadability according to the classification by Dietz (22). The value of 8.08 mm for macadamia nut oil is consistent with that reported by Akhtar et al. (28) The low spreadability is also the reason for adding dimethicone (with a viscosity of 350 cSt) to the formulation, as this will increase the spreadability of the hand cream. It is clear that the three insect fats have a lower spreadability and viscosity. This lack of any correlation with what would be expected according to the trend line is probably due to the presence of a considerable amount of unsaponifiables, as also seen in the TLC results. This demonstrates the difficulty of making generalizations in the presence of compounds other than triglycerides.

Table V  
Viscosity of Insect Fats and Selected Reference Oils

Fat	Viscosity (cP)	Viscosity (cP) (18)
Isopropyl myristate	13.3	4.6
Cyclopentasiloxane	13.3	4.6
Locust oil	20	/
Cricket oil	23.3	/
Octyl stearate	23.3	12.2
C12-C15 alkyl benzoate	25	11.8
Decyl oleate	25	13.5
C8/C10 Triglyceride	36.7	/
BSF oil	41.7	/
Octyldodecanol	60	44.7
Macadamia nut oil	83.3	/
Mink oil	98.3	/
Dimethicone	396.7	90

Table VI  
Spreadability of Insect Fats on Gelatin Films

Species	Spreadability (mm)	Std. Dev.
Cricket	6.07 (8)	0.2
Locust	5.86 (8)	0.2
BSF	6.44 (4)	0.6
Mink	8.76 (4)	0.9
Macadamia nut	8.08 (4)	0.4

The reported values are averages and the number of measurements is given between brackets.

#### PROOF OF PRINCIPLE: A HAND CREAM PREPARATION

A hand cream was prepared to assess the physical stability of the cream and the efficacy of the preservative system when using insect fats. These are two important criteria any cosmetic product should meet before being placed on the market. No perfume was added to monitor any changes in odor more carefully. Various fat concentrations ranging from 1% to 10% (w/w) were tested to evaluate the physical stability and sensorial properties of the cream as a function of fat concentration.

As the fat concentration was increased the color of the cream darkened and the specific fat odor became more pronounced. This was particularly the case at 10% fat, which resulted in a hand cream with undesirable organoleptic properties.

The stability was tested by storing the creams at 5°C, 21°C, and 45°C for 2 mo and monitoring the color, scent, viscosity, pH, and general aspect (i.e., whether or not a phase separation can be observed) at various time points. The 5°C and 45°C conditions represent enhanced aging conditions that allow a prediction of the long-term product stability at ambient conditions. Table VII summarizes the stability results for the cream containing 5% fat, which are representative for all other concentrations tested. The odor and color values at 21°C can also be taken as the initial reference values.

Overall the emulsions remained homogeneous and there was no significant change in the viscosity or pH value of the cream. Only in the case of creams with BSF fat was a slight syneresis observed after 4 wk at 45°C. This suggests that the long-term stability of

Table VII  
Stability of 5% Insect Hand Creams after 2 mo

Cream	Odor			Color			Aspect	Remark
	5°C	21°C	45°C	5°C	21°C	45°C	5°C, 21°C, 45°C	
Locust raw	++	++	++	7	7	8	Homogeneous	Discoloration
Cricket raw	--	-	++	2	3	3	Homogeneous	
BSF raw	-	-	+	3	2	3	Homogeneous	Slight syneresis at 45°C
Cricket decolorized	--	-	-	1	2	2	Homogeneous	
Locust decolorized	++	++	++	6	7	7	Homogeneous	Discoloration
Locust decolorized/ deodorized	--	--	--	6	7	7	Homogeneous	Discoloration
BSF deodorized	+	-	+	3	2	3	Homogeneous	Slight syneresis at 45°C

Color scale: 1 = white to 10 = dark yellow (or green in case of locust fat). Odor scale: -- Negligible smell; - Odor noticeable, but not off-putting; + Odor noticeable and slightly off-putting; ++ Clear, off-putting odor.

these creams may pose an issue. Nevertheless, this can likely be solved by making slight modifications to the cream's composition. The color of the creams was unchanged after 2 mo except for those containing locust fat. Here a discoloration of the cream from greenish to white at the emulsion–air interface was observed. Addition of an antioxidant (0.25% sunflower seed oil extract of rosemary leaf) countered this effect, indicating that the discoloration is due to oxidation, presumably of the chlorophyll present in the fat.

The final preservative efficacy was tested only on a hand cream containing 5% insect fat. This is the standard concentration when using mink or macadamia nut oil. The microbial stability of the cream, or preservative efficacy, was assessed by a challenge test according to the European Pharmacopoeia (21). All creams containing insect fats passed the challenge test according to the A-criteria as all inoculated microorganisms are killed within a certain time and there is no proliferation of microorganisms. This is illustrated in Figure 3, which shows the microorganism reduction plot as a function of time for *P. aeruginosa* and *A. brasiliensis*. The curves for the other microorganisms tested overlap with that of *P. aeruginosa*. This implies that the insect fats do not interfere with the preservative system.

## CONCLUSIONS

Insects have the potential to provide a durable source of biomaterials such as fats. Here we extracted fats from BSE, crickets, and locusts and implemented them in a hand cream

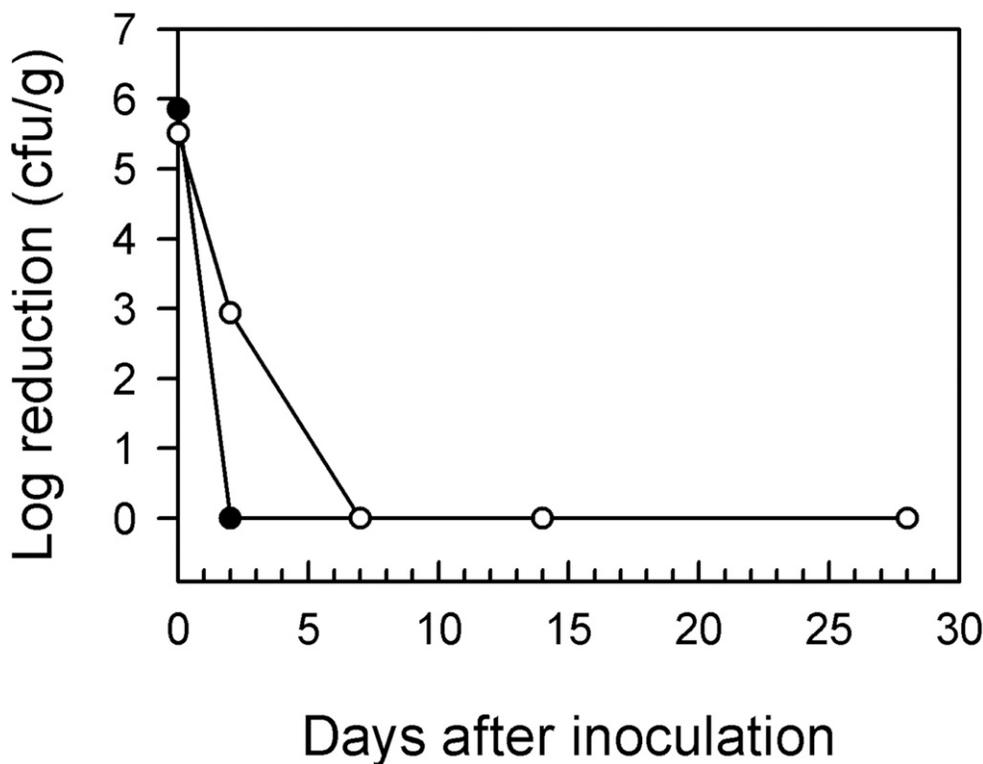


Figure 3. Microorganism reduction in locust hand cream after inoculation according to the European Pharmacopoeia (21). Symbols: *P. aeruginosa* (●) and *A. brasiliensis* (○).

formulation. The data indicate that insect fats contain a large fraction of FFAs and phospholipids which will need to be removed by thorough refining processes to make them more suitable for cosmetics applications. Extraction of insect biomass needs to be performed cost-effectively and green processes (e.g., pressing) should be implemented where possible. Finally, all biomass derived from insects (fats, proteins, and chitin) needs to be valorized in industrial applications. If, in addition, a drastic upscaling of capacity (e.g., stacking) and reduction in breeding costs (e.g., by using waste streams as insect feed) is achieved then industrial implementation of insects becomes feasible and may prove a viable sector in a future circular economy.

As an example of the use of insects in such a circular economy we considered the application of insect fats in a hand cream that serves as a model system for a typical oil-in-water emulsion. Taken together the results of the physicochemical and stability tests, demonstrate that insect fats (especially cricket and the locust, when properly discolored) are suitable for leave-on cosmetic preparations, at least from a physicochemical point of view. BSF fats have a fatty acid profile that is similar to coconut oil and palm kernel oil (29). These oils are frequently used in cosmetics applications e.g., as a starting material for preparation of surfactants (e.g., Amillite GCS-11). Therefore it can be envisaged that BSF fats can be used for similar applications as these plant materials. However, a full toxicological assessment needs to be performed, before considering actual implementation. This also includes an investigation of the potential presence of undesired contaminants such as pesticides and residual solvents.

In conclusion, our data indicate that insects can be implemented as an alternative source for fats that are useable for cosmetic applications. Depending on the fatty acid profile of the insect fats, different applications can be envisaged.

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#### REFERENCES

- (1) O. K. Shortall, S. Raman, and K. Millar, Are plants the new oil? Responsible innovation, biorefining and multipurpose agriculture, *Energy Policy*, **86**, 360–368 (2015).
- (2) E. B. Fitzherbert, M. J. Struebig, A. Morel, F. Danielsen, C. A. Brühl, P. F. Donald, and B. Phalan, How will oil palm expansion affect biodiversity? *Trends Ecol. Evol.*, **23**(10), 538–545 (2008).
- (3) T. M. Fayle, E. C. Turner, and J. L. Snaddon, Oil palm expansion into rain forest greatly reduces ant biodiversity in canopy, epiphytes and leaf-litter, *Basic Appl. Ecol.*, **11**, 337–345 (2010).
- (4) M. J. M. Senior, K. C. Hamer, and S. Bottrell, Trait-dependent declines of species following conversion of tropical forest to oil palm plantations, *Biodivers. Conserv.*, **184**, 414–423 (2013).
- (5) J. A. Foley, N. Ramankutty, K. A. Brauman, E. S. Cassidy, J. S. Gerber, M. Johnston, N. D. Mueller, C. O'Connell, D. K. Ray, P. C. West, C. Balzer, E. M. Bennett, S. R. Carpenter, J. Hill, C. Monfreda, S. Polasky, J. Rockström, J. Sheehan, S. Siebert, D. Tilman, and D. P. M. Zaks, Solutions for a cultivated planet, *Nature*, **478**, 337–342 (2011).
- (6) D. Pimentel, A. Marklein, M. A. Toth, M. N. Karpoff, G. S. Paul, R. McCormack, J. Kyriazis, and T. Krueger, Food versus biofuels: environmental and economic costs, *Hum. Ecol.*, **37**(1), 1–12 (2009).
- (7) A. van Huis, J. Van Itterbeeck, H. Klunder, E. Mertens, A. Halloran, G. Muir, and P. Vantomme, FAO Forestry Paper 171. Edible insects: future prospects for food and feed security. ISBN 978-92-5-107595 (2013).

- (8) S. Diener, C. Zurbrügg, F. R. Gutiérrez, D. H. Nguyen, A. Morel, T. Koottatep, and K. Tockner, "Black soldier fly larvae for organic waste treatment—prospects and constraints," in Proceedings of the Waste-Safe 2011—2nd International Conference on Solid Waste Management in the Developing Countries. Khulna, Bangladesh, ISBN: 978-984-33-2705-5 (2011).
- (9) H. Čičková, L. G. Newton, C. R. Lacy, and M. Kozánek, The use of fly larvae for organic waste treatment, *Waste Manage.*, **35**, 68–80 (2015).
- (10) Q. Li, L. Zheng, Y. Hou, S. Yang, and Z. Yu, Insect fat, a promising resource for biodiesel, *J Phylogenetics Evol Biol* S2:001. doi:10.4172/2157-7463.S2-001. (2011).
- (11) L. Zheng, Q. Li, J. Zhang, and Z. Yu, Double the biodiesel yield: rearing black soldier fly larvae, *Hermetia illucens*, on solid residual fraction of restaurant waste after grease extraction for biodiesel production, *Renewable Energy*, **41**, 75–79 (2012).
- (12) H. K. V. Prashanth and R. N. Tharanathan, Chitin/chitosan: modifications and their unlimited application potential—an overview, *Trends Food Sci. Technol.*, **18**, 117–131 (2007).
- (13) G. R. DeFoliart, Insect fatty acids: similar to those of poultry and fish in their degree of unsaturation, but higher in the polyunsaturates, *Food Insects Newsl.*, **4**, 1–4 (1991).
- (14) H. A. C. Le Poole, Natural Oils and Fats Multifunctional Ingredients for Skin Care (Dekker, Wormerveer, The Netherlands, 1994), pp. 47–56.
- (15) G. N. Stamatias, J. de Sterke, M. Hauser, O. von Stetten, and A. van der Pol, Lipid uptake and skin occlusion following topical application of oils on adult and infant skin, *J. Dermatol. Sci.*, **50**(2), 135–142 (2008).
- (16) J. Alander, "Cosmetics" in Vegetable Oils and Fats (Aarhuskarlshamn, Karlshamn, Sweden, 2007), pp. 214–234.
- (17) FAVV Gemeenschappelijk advies SciCom 14-2014 en HGR Nr. 9160, Food safety aspects of insects intended for human consumption (SHC 9160) (report of the FASFC - Sci Com 2014/04) (2014).
- (18) D. C. Sheppard, J. K. Tomberlin, J. A. Joyce, B. C. Kiser, and S. M. Sumner, Rearing methods for the black soldier fly (Diptera: Stratiomyidae), *J. Med. Entomol.*, **39**(4), 695–698 (2002).
- (19) M. A. Kaluzny, L. A. Duncan, M. V. Merritt, and D. E. Epps, Rapid separation of lipid classes in high yield and purity using bounded phase columns, *J. Lipid Res.*, **26**, 135–140 (1985).
- (20) S. O'Keefe, "Fat characterization," in Food Analysis, 4th Ed. (Springer, New York, 2003), pp. 239–260.
- (21) European Pharmacopoeia 7.0, Efficacy of antimicrobial preservation. In: Europeann Pharmacopoeia volume 7.0, pp. 505–506 (01/2011:50103).
- (22) T. Dietz, Basic properties of cosmetic oils and their relevance to emulsion preparations, *SÖFWJ.*, **125**, 7 (1999).
- (23) F. R. van de Voort, J. Sedman, and J. Russin, Lipid analysis by vibrational spectroscopy, *Eur. J. Lipid Sci. Technol.*, **103**, 815–840 (2001).
- (24) D. A. Tzompa-Sosa, L. Yi, H. J. F. van Valenberg, M. A. J. S. Boekel, and C. M. M. Lakemond, Insect lipid profile: aqueous versus organic solvent-based extraction methods, *Food Res. Int.*, **62**, 1087–1094 (2004).
- (25) A. Zielińska and I. Nowak, Fatty acids in vegetable oils and their importance in cosmetic industry, *Chemia*, **68**(2), 103–110 (2004).
- (26) P. G. Green, R. H. Guy, and J. Hadgraft, In vitro and in vivo enhancement of skin permeation with oleic and lauric acids, *Int. J. Pharm.*, **48**, 103–111 (1988).
- (27) X. Liu, X. Chenn, H. Wang, Q. Yang, K. Ur Rehman, W. Li, M. Cai, Q. Li, L. Mazza, J. Zhang, Z. Yu, and L. Zheng, Dynamic changes of nutrient composition throughout the entire life cycle of black soldier fly, *PLOS One*, **12**(8), e0182601 (2017).
- (28) N. Akhtar, Q. Adnan, M. Ahmad, A. Madni, and M. S. Bakhsh, Evaluation of basic properties of macadamia nut oil, *Gomal Univ. J. Res.*, **22**, 21–27 (2006).
- (29) V. Dubois, S. Breton, M. Linder, J. Fanni, and M. Parmentier, Fatty acid profiles of 80 vegetable oils with regard to their nutritional potential, *Eur. J. Lipid Sci. Technol.*, **109**, 710–730 (2007).